Noninvasive Prenatal DNA Testing: The Vanguard of Genomic Medicine

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Keywords

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Abstract

Noninvasive prenatal DNA testing is the vanguard of genomic medicine. In only four years, this screening test has revolutionized prenatal care globally and opened up new prospects for personalized medicine for the fetus. There are widespread implications for increasing the scope of human genetic variation that can be detected before birth, and for discovering more about maternofetal and placental biology. These include an urgent need to develop pretest education for all pregnant women and consistent post-test management recommendations for those with discordant test results. The reduction in invasive testing has had downstream effects on specialist training and caused many countries to re-examine their national approaches to prenatal screening. Finally, the accumulating datasets of genomic information on pregnant women and their fetuses raise ethical issues regarding consent for future data mining and intellectual property.

INTRODUCTION

CVS: chorionic villus sampling

CFTS: combined first-trimester screening

The current era of genomic medicine is rapidly advancing our understanding of disease and creating novel personalized approaches to therapy (1, 2). Despite the high expectations from the scientific and general communities, there are relatively few large-scale examples of genomic medicine transforming routine clinical care. One notable example is the analysis of circulating DNA fragments in the plasma of pregnant women for the noninvasive detection of fetal genetic abnormalities such as trisomy 21. In only four years, this screening test has revolutionized prenatal care globally and opened up new prospects for personalized medicine for the fetus (3).

A detailed review of the different technologies used to analyze maternal plasma DNA was published last year in this journal (4). The wider implications of these stunning technical advances for the delivery of prenatal care also demand attention. In this review, we first address the important shifts in the traditional concepts in prenatal screening brought about by DNA sequencing technology, and then review its broader impact on healthcare delivery and society. Because of the rapid incorporation of maternal plasma cell-free DNA analysis into care, and the large number of samples tested to date (more than two million), prenatal genomics is, in a sense, on the vanguard of genomic medicine.

BACKGROUND: PRENATAL SCREENING FOR CHROMOSOME ABNORMALITIES

Trisomy 21 is the most common chromosome abnormality to affect liveborn children. Historically, it has therefore been the major focus of prenatal screening. Advanced maternal age is the most important risk factor for this condition. Women aged 40 years or more have an approximately 1% risk of delivering a liveborn infant with trisomy 21 (Down syndrome). Prenatal diagnosis is possible by performing cytogenetic or DNA analysis of fetal cells obtained by amniocentesis or chorionic villus sampling (CVS), but owing to the small risk of procedure-related miscarriage, screening tests are usually offered to refine the risk of an affected pregnancy prior to consideration of invasive diagnostic testing. Conventionally, only women with a "high-risk" result (using locally defined thresholds) are offered diagnostic testing. In many developed countries, voluntary prenatal screening for fetal chromosome abnormalities is a standard component of maternity care (5, 6).

Screening approaches for Down syndrome have evolved slowly over the past four decades, from the sole criterion of advanced maternal age to second-trimester serum screening with multiple analytes produced by the fetus or placenta. Until recently, the test with the highest detection and lowest false-positive rates was combined first-trimester screening (CFTS), consisting of ultrasound examination and serum markers. The combination of maternal serum levels of free β-hCG and pregnancy-associated plasma protein-A (PAPP-A), with measurement of the fetal nuchal translucency by ultrasound examination at 11–13 weeks of gestation, resulted in trisomy 21 detection rates of 85–90% for a false positive rate of 5% (7, 8). The later introduction of the fetal nasal bone measurement as an additional sonographic marker further reduced the false positive rate to approximately 3%. These incremental improvements in screening are reflected in population-based increases in diagnostic yield from invasive testing (9).

However, standard prenatal practice was disrupted in 2011 by the introduction of cell-free DNA-based screening into clinical care by industry. The scientific basis of the test dates back to 1997, when it was first reported that cell-free DNA from the fetus could be detected in maternal plasma (10). It was subsequently established that the specific source of the circulating DNA was cytotrophoblast (11). Approximately 10–15% of the cell-free DNA in maternal plasma originates from the placenta (12); the remainder derives from maternal hematopoietic cells undergoing

apoptosis (13) and, in obese women, necrosis and apoptosis of adipocytes and stromal cells (14). A fetus affected with Down syndrome will release more DNA fragments from chromosome 21 than an unaffected fetus because of the presence of trisomy in the trophoblast cells of the placenta. DNA sequencing and counting allow the very precise determination of the proportion of chromosome 21–derived DNA fragments in maternal plasma as compared to a reference value. If there are significantly more fragments than expected, then this suggests that the fetus is aneuploid. Applying this counting principle, and using reasonably cost-effective DNA sequencing techniques, noninvasive screening for trisomy 21 was rapidly shown to have excellent clinical performance, with a detection rate of >99% and false-positive rate of <1 in 1,000 (15). Testing was then extended to the detection of trisomies 18 and 13, thus providing coverage for the three most common fetal autosomal aneuploidies that result in a live birth.

PPV: positive predictive value

The improved technical performance of maternal plasma DNA analysis did not mean, however, that integration into clinical care was uniform or without controversy. In the early days of implementation, there were many misconceptions that noninvasive prenatal testing (NIPT) was diagnostic and that confirmation of aneuploidy-positive results was not necessary. Shortly after maternal plasma cell-free DNA testing became clinically available, reports began to appear with titles such as, "Is It Time to Sound an Alarm about False-Positive Cell-Free DNA Testing for Fetal Aneuploidy?" (16). Although the very low false-positive rate of NIPT for trisomy 21 does mean a very high risk of an affected fetus after an abnormal NIPT result [i.e., NIPT has a high positive predictive value (PPV)], NIPT is not diagnostic. All professional societies globally recommend that high-risk results be confirmed with diagnostic testing (17, 18). Despite this, in one study, as many as 6% of pregnant women terminated their pregnancies without obtaining a follow-up diagnostic test because of anxiety generated by a positive result (19).

It is also important to recognize that PPVs correlate with the background prevalence of the disease, as this alters the ratio of true-positive (TP) to false-positive (FP) results [PPV = TP/(TP + FP)]. Therefore, for the same sensitivity and specificity, the PPV of NIPT will be lower in low-risk women, although it still outperforms CFTS. In a clinical study that compared NIPT with standard screening in a low-risk population (average maternal age 30 years), the PPV for trisomy 21 was 45.5% with NIPT versus 4.2% with standard screening (20).

Another new issue encountered in the early clinical implementation of NIPT was grappling with the management of women who did not receive a result from their blood sample. A small proportion of samples submitted for NIPT will have a test failure, most commonly due to a low amount of placental cell-free DNA in maternal blood, or low "fetal fraction" [fetal fraction = placental DNA/(placental DNA + maternal DNA)]. Most laboratories require a minimum fetal fraction of 2–4% for a reliable result (21). Approximately half of women with a "no call" result will obtain a successful result on repeat sampling, but those who do not may lose the opportunity to access CFTS if their gestation has advanced past 13 weeks 6 days. In addition, pregnancies with very low fetal fractions may actually be at higher risk of certain abnormalities, such as trisomy 18 or triploidy, and therefore require specialized management (22).

In the wider context, implementing NIPT into existing publicly funded healthcare models has been challenging, given its initial development by private commercial laboratories and the relatively high cost of the test (approximately $US 1,500–2,000 at the time of introduction). Various strategies for implementation of maternal plasma cell-free DNA analysis into prenatal care have been proposed, each with its advantages and disadvantages for detection rates and costs (**Figure 1**).

In addition to the debate around the optimal method of incorporating a high-performing but expensive technology into a public health model, other controversies related to the implementation of genomic testing on a broad scale ensued. These included (*a*) the potential to expand screening

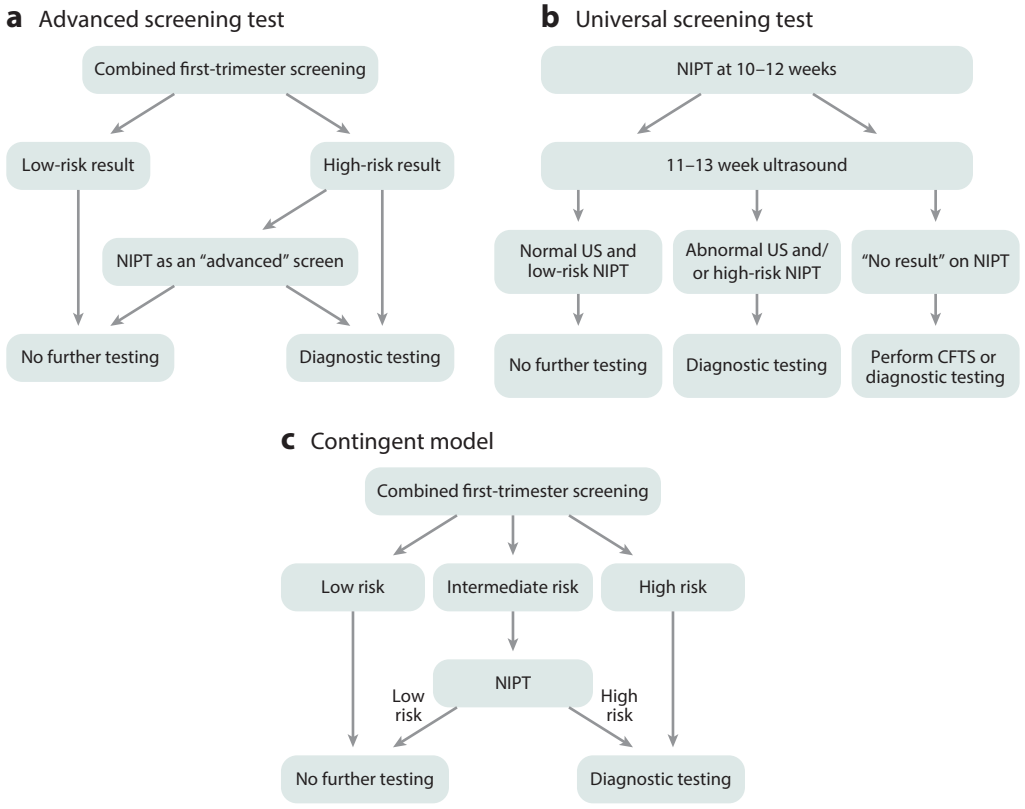

Figure 1

Models of incorporating noninvasive prenatal testing with existing screening methods. (*a*) "Advanced" or "secondary" screening. Noninvasive prenatal testing (NIPT) is offered to women at "high risk" based on prior conventional screening tests. This method reduces invasive testing rates and has a variable effect on the overall detection rate of trisomy 21. The UK National Screening Committee has recommended evaluating the introduction of this model within the National Health Service (**http://legacy.screening.nhs.uk/fetalanomalies**). (*b*) "Universal primary screening" provides NIPT and a 12-week ultrasound (US) examination to all pregnant women. This provides the maximum detection rate of fetal aneuploidy and provides early detection of structural abnormalities but is the most expensive model (23). Abbreviation: CFTS, combined first-trimester screening. (*c*) The "contingent model"of screening offers NIPT only to women with a locally defined "intermediate risk" of aneuploidy after combined first-trimester screening (e.g., risk between 1:100 and 1:1,000). This model is less costly than the universal model and improves aneuploidy detection over the "advanced screening test" model because a larger group of women is offered NIPT (24).

beyond the common autosomal trisomies (13, 18, 21) and (*b*) the detection of secondary findings with new implications for maternal health.

CLINICAL IMPACT ON THE PARADIGM OF PRENATAL SCREENING

Expanding the Scope of Prenatal Screening

The use of maternal plasma cell-free DNA analysis instead of serum analytes or nuchal-translucency measurement to screen for the common autosomal aneuploidies was a change in technology, but it did not fundamentally change the conditions for which screening was being provided. Screening based on cell-free DNA, however, represents a direct measure of chromosome dosage, as opposed to indirect measures of proteins or hormones. Thus, the entire fetal genome

can be analyzed, providing the option of noninvasively prenatally detecting conditions for which no prior screening protocols exist. Examples of these conditions include relatively common but prenatally asymptomatic sex chromosome aneuploidies such as Klinefelter syndrome (47, XXY) and subchromosomal deletions including extremely rare disorders such as Langer-Giedion and Jacobsen syndromes. Clinical experience and professional recommendations continue to evolve with regard to screening for subchromosomal microdeletions and microduplications, but some broad principles are summarized here.

SCA: sex chromosome aneuploidy

CPM: confined placental mosaicism

CNV: copy-number variant

Sex chromosome aneuploidies. Testing for sex chromosome aneuploidies (SCAs) was first added as a screening option in 2012, initially for monosomy X alone (25). This was quickly followed by testing for other SCAs, including 47, XXX; 47, XXY; and 47, XYY, which are frequently mild or asymptomatic and do not meet the classical criteria for population-based screening. The performance of NIPT for the X and Y chromosomes is, however, not as accurate as it is for chromosome 21. A recent meta-analysis estimated the sensitivity for detection of monosomy X at 90.3% and the false-positive rate of 0.23%, as compared with >99.2 and 0.09%, respectively, for trisomy 21 (26). For the other SCAs, the detection rate was estimated at 93.0% and the false-positive rate at 0.14%. SCAs that include the Y chromosome are generally more accurate because the mother does not have a Y, which makes it easy to distinguish in maternal plasma. As the false-positive results for each additional condition have a cumulative effect, clinical experience indicates that ~1% of samples will return a high-risk result for an SCA, somewhat reducing the gains in specificity from trisomy 21 screening (27).

There are several reasons for false-positive SCA results. These include confined placental mosaicism (CPM), demise of a cotwin with monosomy X or triple X, or maternal constitutional or somatic X chromosome numerical abnormalities. CPM for monosomy X is a relatively common event, occurring in 59% of pregnancies with evidence of monosomy X on CVS (28). On the other hand, undiagnosed maternal SCA mosaicism is also more common than previously thought. Wang and colleagues (29) reported that 16/181 (8.6%) of the women with high-risk NIPT results involving the X chromosome had an underlying maternal cause for the result (e.g., 47, XXX). This suggests that the clinical spectrum of SCAs is broader than previously realized and that women with SCAs can be fertile.

Another underrecognized maternal phenomenon has also come to light with growing clinical experience. Bianchi et al. (27) demonstrated that the average maternal age in false-positive cases of monosomy X was statistically significantly higher than the mean for true-positive cases (36.7 versus 31.7 years, $p < 0.001$). This appears to be explained by the age-related somatic loss of one X chromosome in the peripheral blood, leading to low-level somatic mosaicism in some women (30).

Microdeletions and microduplications. Following the publication of several proof-of-principle studies (31–39), microdeletion testing became clinically available in 2013. The current test options offered by commercial laboratories in the United States include 5p– (cri du chat), 22q11.2– (Di George syndrome), 15q– (Prader-Willi/Angelman syndrome), 4p– (Wolf-Hirschhorn syndrome), 11q– (Jacobsen syndrome), 8q– (Langer-Giedion syndrome), and 1p36–. There are several rationales for expanding screening to include rare subchromosomal abnormalities with severe phenotypes: (*a*) 1.7% of sonographically normal fetuses with a normal G-banded karyotype have a clinically significant copy-number variant (CNV) on microarray analysis (40); (*b*) using NIPT to detect only trisomies 13, 18, and 21 in high-risk women would miss 16.9% of the total cytogenetic abnormalities currently detected with diagnostic testing (41); and (*c*) the risk of fetal microdeletions and microduplications is independent of maternal age, and for pregnant women under age 40 they are collectively more common than trisomy 21.

After two years of clinical laboratory experience testing for microdeletions, several reports on clinical test performance were published (42, 43). As of mid-2016, the true sensitivities and negative predictive values of microdeletion screening in clinical practice are still uncertain because the current studies are limited by incomplete outcome data, variation in the definitions of confirmed cases, and differences in methods of calculating test performance (44). Most of the microdeletion syndromes are very rare. It is unlikely, therefore, that a prospective clinical validation study with complete outcome information can or will ever be performed.

The rarity of each microdeletion syndrome also means that screening for these conditions inherently has lower PPVs than screening for the common autosomal trisomies. The risk of an affected pregnancy given a positive test result for 22q11.2 deletion is likely to be <5% in low-risk women with normal fetal ultrasound findings. Conversely, the high negative predictive value of NIPT for microdeletions (i.e., the chance of an unaffected pregnancy given a low-risk result) reflects the rarity of each of the syndromes more than test performance per se (45).

Furthermore, the sensitivity of NIPT for microdeletions is inherently inferior to whole-chromosome aneuploidy, as it does not detect the atypical forms of the deletions (22q11.2 deletions <3 Mb, or 15q– phenotypes caused by uniparental disomy). The strong reservations about the expected clinical performance of microdeletion screening were reflected in the 2016 practice bulletin from the American College of Obstetricians and Gynecologists and the Society for Maternal-Fetal Medicine (46, p. 981), which stated that "cell-free DNA screening tests for microdeletions have not been validated clinically and are not recommended at this time."

Despite the careful opinions of professional bodies, the boundaries of prenatal screening continue to be pushed by the possibilities afforded by technology. Inevitably, NIPT has expanded beyond selected whole-chromosome aneuploidy to genome-wide assessment at a similar resolution to a conventional G-banded karyotype. Lefkowitz et al. (47) reported on genome-wide CNVs in 1,166 high-risk pregnancies at a depth of 32 million reads per sample. The study samples were assessed for trisomies 13, 18, and 21, SCAs, CNVs >7 Mb, and selected deletions <7 Mb. There were 42 true-positive, 1 false-positive, and 1 false-negative results for CNV detection, producing an estimated sensitivity of 97.7% (95% CI 86.2–99.9) and specificity of 99.9% (99.4–100). This relentless progress toward the comprehensive, noninvasive assessment of the fetal genome provides many exciting opportunities and simultaneous challenges for the field of prenatal screening.

Unintended Consequences and New Biological Insights from Noninvasive Prenatal Testing

Discordant positive results have provided new insights into maternofetal and placental biology. Cell-free DNA in maternal plasma is a biomarker of placental health and disease (48). It is therefore not surprising that a major reason for discordant NIPT results is the presence of CPM, which from CVS data was thought to occur in 1–2% of viable gestations (49). Furthermore, the rates of CPM are higher in specific aneuploidies. By comparing CVS and amniocentesis karyotypes in women who had both procedures, Grati and colleagues (28) showed that the rates of CPM were much higher for trisomy 13 (22% of cases) and monosomy X (59%) than for trisomies 21 (2%) and 18 (4%).

Another major reason for discordant test results is the presence of a deceased twin (50). This is a consideration when there is a singleton female fetus and Y chromosomal DNA is detected. Using single-nucleotide polymorphism (SNP) technology, Curnow et al. found haplotypes in maternal plasma that were not shared by the mother or surviving fetus in 130 of >30,000 (0.43%) consecutive clinical cases. These authors suggested that the additional DNA fragments originated in an undetected twin, a demised twin, or a triploid fetus.

A third reason for discordant results is that in pregnancy, the maternal and fetal DNA fragments circulating in maternal plasma are simultaneously analyzed. This creates an opportunity to detect secondary, or incidental, findings in the mother. Although it is recommended that pretest counseling include a specific discussion of the possibility of finding maternal abnormalities (51), in practice this does not always occur (52). Following NIPT, a variety of maternal secondary findings have been reported in the literature, including sex chromosome abnormalities (described above), mosaicism for autosomal aneuploidy (53), microdeletions such as 22q11.2 (42), and large CNVs (54). In one particularly ethically troubling case, a seemingly false-positive NIPT result for trisomy 21 was shown to be due to a familial partial duplication of 21q21.1 (55). This was outside of the Down syndrome critical region, but included the gene *amyloid precursor protein* (*APP*). As a result of her decision to undergo NIPT for fetal aneuploidy, the pregnant proband and two others (her identical twin sister and her fetus) were inadvertently identified to be at risk for early-onset Alzheimer disease.

Prenatal genomic testing has also identified maternal medical conditions such as vitamin B12 deficiency (56) and uterine fibroids (57). The cases that have received the most attention, however, have been the maternal incidental findings of cancer (58–61). Whereas in oncology there is much discussion about the future possibilities of liquid biopsy and noninvasive detection of malignancy, prenatal testing has already shown that it can be done. In cases of maternal malignancy, however, initial test results may be confusing and reported as multiple aneuploidies or as test failures (53, 61). This is due to the fact that tumors shed large amounts of DNA fragments into the maternal circulation, resulting in genome-wide imbalance, which disrupts the expected ratios of test to reference chromosomes. Cancer during pregnancy is thought to be rare; the results of prenatal genomic testing are challenging this conventional wisdom. Furthermore, the ability to detect maternal malignancy is outpacing the development of evidence-based recommendations for further diagnostic work-up and treatment.

HEALTH-SECTOR AND SOCIETAL IMPACT OF NONINVASIVE PRENATAL TESTING

Rapid Global Dissemination

The speed at which NIPT has been disseminated around the world is unprecedented. By the end of 2014, various companies had expanded the test to market in at least 61 countries (62). The vast majority of early testing was paid for by pregnant women themselves, demonstrating the huge appeal of a highly accurate screening test available from 10 weeks of gestation onward. However, the manner in which NIPT was incorporated into clinical care was highly influenced by local factors, such as the structure of existing screening programs, average patient income, availability of private insurance coverage, and cultural attitudes to prenatal diagnosis. Within a year of NIPT's introduction in the United States, private health insurers began to offer coverage for high-risk women as costs declined and the benefits of reducing unnecessary invasive testing and improving detection rates became evident.

Elsewhere, in countries with established government-funded prenatal screening programs, NIPT forced a complete re-examination of health policies. Several countries with nationalized health services have conducted their own prospective clinical trials of NIPT. In the United Kingdom, the Reliable Accurate Prenatal non-Invasive Diagnosis (RAPID) project offered NIPT to women whose risk of trisomy 21, 18, or 13 was 1:1,000 or higher after a primary screening test (most commonly CFTS). After considering the results of this study, the UK National Screening Program recommended introducing government-funded NIPT for women with aneuploidy risks

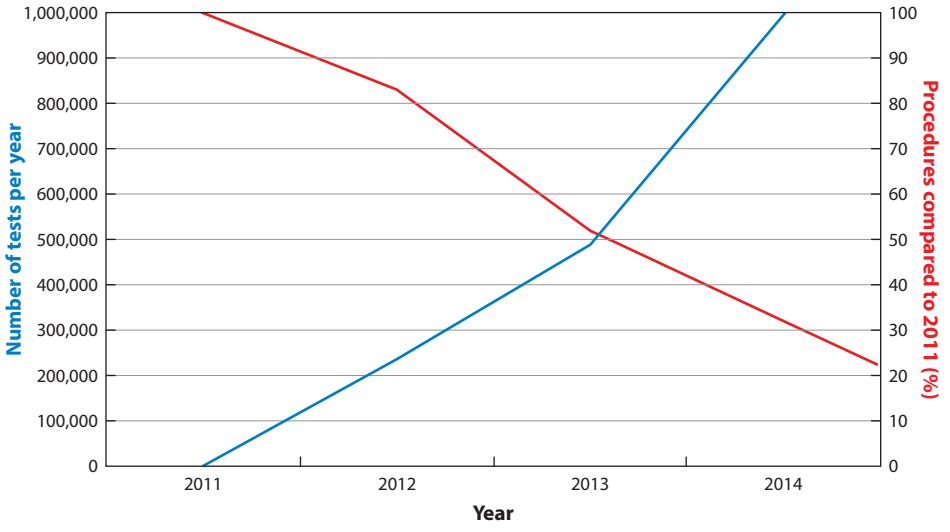

Figure 2

Decline in invasive prenatal diagnosis in association with increased uptake of noninvasive prenatal testing (NIPT). Numbers of NIPTs obtained from publications and/or public presentations as reported by Genome Web from BGI, Berry Genomics, Verinata, Sequenom, and Natera. After 2014, testing was licensed to additional laboratories all over the world. This makes it impossible to calculate the current volume of tests being performed. The decline in the number of procedures was estimated from References 65–68. Figure created by Lillian Zwemer, PhD.

>1:150 as part of the National Health Service (NHS) Fetal Anomaly Screening Program (63). Along similar lines, the Netherlands and Canada have commissioned publicly funded evaluations of NIPT into clinical practice via the TRIDENT study (64) and PEGASUS trials, respectively (**http://pegasus-pegase.ca/pegasus**). It therefore appears likely that maternal plasma DNA analysis will become a component of several government-funded screening programs, particularly as sequencing costs continue to decline and the technology disseminates beyond commercial laboratories.

Reduction in Invasive Prenatal Diagnosis

The very high specificity of NIPT has significantly reduced the proportion of women being categorized as "high-risk" following screening. This has consequently reduced the numbers of women who are offered follow-up diagnostic testing. As a result of this, the numbers of amniocenteses and CVS procedures being performed have dropped dramatically (65–68) (**Figure 2**). When NIPT is offered as an advanced screen after CFTS, one in three invasive tests yields a confirmed diagnosis of trisomy 21. This is extremely efficient compared with CFTS, where, due to the higher false-positive rate, >10 invasive tests are performed for each fetus with trisomy 21 detected (63). In the United Kingdom, it has been estimated that if NIPT were offered to all women with a risk of >1:150, there would be 4,870 fewer invasive diagnostic procedures per annum compared to the current strategy of CFTS alone.

Although the decline in invasive testing is clearly one of the major successes of NIPT, it has created an urgent issue to be addressed within specialist training. Complication rates of invasive testing are inversely related to operator and center procedural volume (69, 70). It appears inevitable that centralization of procedures will occur, with fewer specialists capable of performing

these procedures (71). Ongoing audit and quality assurance are now a priority for centers that offer prenatal diagnosis. The obstetric specialty is beginning to recognize the importance of innovation and rigor in training the new generation of proceduralists, such as developing simulation models, and competency-based, rather than volume-based, accreditation (72). The cytogenetics and molecular diagnostic laboratories are also affected by the reduction in prenatal diagnostic samples; this too affects competency and quality assurance (73).

Challenging Barriers to Prenatal Screening

NIPT has also had an impact in countries where prenatal screening has not been previously widely offered for cultural and religious reasons. As an example, for many years, Japanese physicians were not required to provide information on prenatal screening to pregnant women. However, the commercial availability of NIPT initially caused a wave of medical tourism in Hawaii and Guam because Japanese women were seeking to obtain NIPT.

This caused Japanese practitioners to re-examine the role of prenatal screening and assess NIPT within a regulated research setting. Members of the government-supported Japanese NIPT consortium (74, p. 334) concluded that "as the cost of NIPT decreases dramatically in the near future, NIPT will become a standard screening method for pregnant women who request testing for fetal chromosomal abnormalities." The consortium also stated that it was "too late to establish a screening strategy that includes maternal serum or ultrasound markers in Japan," thus indicating a willingness to implement NIPT without developing a CFTS program (74). A similar "leapfrogging" approach of embracing the newest technology has occurred in the Netherlands, where historically there was low uptake of prenatal serum screening for aneuploidy (64).

Impact on Other Components of Clinical Care

Most of the research focus to date has been on improving the technical performance of NIPT, e.g., improving sensitivities and specificities. There has been less attention to developing the other components of clinical care to adjust to patient interest in NIPT. A demand exists for improving the genetic education of both practitioners and the general public. There is also a growing need for associated services such as genetic counseling, interpretation of genomic data, disability services, and safe, accessible abortion.

It is important to recognize that to date, there is no evidence that the introduction of NIPT has led to an increase in termination rates for fetal abnormalities; many women wish to access screening with or without diagnostic confirmation for information and preparation. In a large study from the United Kingdom, in which NIPT was offered contingent on results from CFTS, 91.5% of pregnancies with trisomy 21 were diagnosed and 32% of these resulted in live births (24). These findings are consistent with termination rates in the United States after prenatal diagnosis of trisomy 21 in the pre-NIPT era (67%) (75).

The availability and practice of termination of pregnancy for fetal anomalies may also influence societal attitudes to disability. Some stigma associated with intellectual disability is present in all ethnic groups but is inversely proportional to a person's contact with and awareness of affected individuals (76). There is a concern that reduced numbers of individuals with Down syndrome in the community will lead to decreased social acceptance and fewer disability services.

Patient Attitudes/Engagement as Stakeholders in the Genomic Era

The positive attitudes toward receiving genomic information that have been observed in adult medicine (77) may not translate to the prenatal setting, where it is frequently difficult to predict

the future phenotype of the child. In the context of an uncertain prognosis or a wide spectrum of possible postnatal outcomes, fetal genomic information may lead to maternal anxiety and decisional conflict, resulting in the problem of "toxic knowledge" (78). In a recent international policy statement, the medical profession has recognized the special need for responsible innovation in prenatal testing (79). There also needs to be better pretest counseling as part of the consent process (51), and more support for communicating secondary findings that may have implications for multiple family members (80).

One of the major concerns with ensuring ethical use of the information obtained by NIPT is its potential to contribute to the practice of sex-selective abortion in countries where there is a strong preference for sons and a heavily skewed sex ratio at birth. NIPT allows fetal sex to be determined earlier in gestation than by ultrasound examination. This has led to reservations about providing widespread clearance for its use in some countries, such as India, that currently legislate against providing information on fetal sex (81).

Finally, the issue of ownership of genomic information is becoming blurred as commercial laboratories reanalyze large genomic datasets accumulated through clinical testing for new purposes. Plasma samples collected from pregnant women for NIPT have been used to derive population estimates of maternal CNVs (82), and mapping of SNPs and genetic variants in the Han Chinese population (83). Long-term data storage may mean that consent processes for use of prenatal genomic information will need to include the capacity to "reconsent" for new research questions or methods, or when the fetus/newborn becomes an adult (84).

CONCLUSIONS

Noninvasive prenatal DNA testing is the vanguard of genomic medicine. The fast pace of clinical translation has been made possible by the huge investment in research and development by private industry. The test has disrupted the 40-year-old prenatal screening paradigm that had been developed, validated, and implemented by academic laboratories and university-based specialists. Its impact is still being felt and is yet to be truly comprehended. There are widespread implications for increasing the scope of human genetic variation that can be detected before birth, and also for discovering more about maternofetal and placental biology. There is an urgent need to develop consistent posttest management recommendations for pregnant women with discordant test results. The reduction in invasive testing and classical cytogenetics has caused different countries to re-examine their national approaches to prenatal screening in terms of their cost effectiveness and cultural aspects. In some settings, maternal plasma cell-free DNA analysis may improve access to screening and may result in opening dialogue on other important issues, such as access to services for children with physical and intellectual disabilities and safe termination of pregnancy. Finally, the large accumulating datasets of genomic information on pregnant women and their fetuses raise ethical issues regarding consent for future data mining and intellectual property as newer tests are developed. Although many other areas of genomic medicine are theoretically exploring similar concepts, it is in the prenatal application that these issues are being dealt with in reality.

SUMMARY: WHY IS NONINVASIVE PRENATAL TESTING AT THE VANGUARD OF GENOMIC MEDICINE?

Wide-scale integration of maternal plasma sequencing into care has resulted in many clinical care changes and magnified some ethical and social issues in prenatal screening.

- Industry, rather than academic laboratories, has driven innovation.
- Expanding test menus have changed the paradigms for prenatal screening.

- Some tests have been introduced before their clinical utility has been established.
- The reduction in invasive procedures has downstream consequences for specialist training.
- Simultaneous sequencing of maternal and fetal DNA creates the potential to detect unexpected maternal genomic abnormalities.
- Sequencing-based screening has increased the complexity of pre- and posttest counseling, exposing an urgent need for professional education and clinical guidance in prenatal genetics and genomics.
- "Leapfrogging" of technology has occurred in some places, such as Japan and the Netherlands.

DISCLOSURE STATEMENT

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